SOME NOTES ON THE ABUNDANCE, ENVIRONMENT AND NUTRITION OF SIPUNCULUS NUDUS L. (SIPUNCULOIDEA) AT MORGAT, BRITTANY

by

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Résumé

Des spécimens de Sipunculus nudus ont été trouvés en nombre considérable sur la plage de sable de Morgat (Finistère), au cours des mois d'août et septembre 1960. Sur une superficie de 100 m/10 m et en deux récoltes, ont été récoltés 4,6 ± 0,98 sipunculides au m². Le sable dans lequel vivent ces animaux est très fin et contient très peu de vase, une haute teneur en carbonate et une faible teneur en azote. La composition granulométrique et le pourcentage en carbonate du contenu intestinal et du sable dans lequel vivait l'animal était approximativement les mêmes. La teneur en azote dans le contenu intestinal était environ deux fois celle du sable. Il était difficile d'estimer l'importance de ce résultat, la teneur en azote des sucs digestifs et du mucus intestinal n'étant pas connue. Quelques squelettes de diatomées furent trouvés dans le contenu de l'intestin antérieur. La plupart d'entre eux étaient vides de substance organique et l'étaient sans doute déjà lors de l'ingestion. Dans ces conditions, il est difficile de dire quelle part importante jouent les diatomées dans la nourriture de S. nudus à Morgat. Les particules de carbonate contenues dans le sable ingéré n'étaient pas décomposées dans le tube digestif. Le pH du contenu intestinal était de 7,5 à 8,2. L'eau était absorbée à partir du contenu intestinal lors de son passage le long du canal alimentaire.

These notes deal chiefly with some problems concerning Sipunculus nudus L. and its environment. S. nudus is a sand-dwelling marine invertebrate that is abundant at a number of places along the Atlantic coast of France. The work was carried out from the Biological Station at Roscoff during August and September, 1960.

MATERIAL

The specimens of *S. nudus* were obtained from the sandy beach at Morgat in the Bay of Douarnenez, Brittany, France. They were collected from an area of the beach about 100 m long and 10 m wide that is situated mid-way between the end of the mole and the divingtower and that is uncovered at the low spring-tides during summer. The worms were found 10-30 cm below the surface of the sand and

CAHIERS DE BIOLOGIE MARINE Tome III - 1962 - pp. 183-190 were dug up with a fork. At Roscoff they were kept in large aquaria containing sand and running sea-water. Many animals after remaining in the aquaria for three months appeared healthy and active.

EXPERIMENTS AND RESULTS

Abundance of the animals.

On two occasions separated by an interval of a month experiments were conducted to find out how abundant the animals were in the area under investigation. In order to do this a wooden frame with inside measurements of 1 m × 1 m was cast at random at 3 m intervals as one walked along the middle of the 100 m strip. The sand within each square was then dug to a depth of about 40 cm and the sipunculids seen were collected and counted. Animals less than 6 cm long in the contracted state were rarely obtained. On the first occasion 152 animals were collected from 32 casts and on the second 135 from 30 casts. The results gave a mean value of 4.6 ± 0.98 sipunculids per square metre. As the beach is about 1000 m long and since sipunculids were found at any spot along the beach at a tide-level corresponding with that of the selected area, it appears that the animals occured at Morgat in great numbers during the time that the study was made. To what extent the animals occured below the level of low-tide was not determined. The sea-urchin, Echinocardium cordatum Forbes, was often dug up while collecting the sipunculids.

Granulometric composition of the sand and of the intestinal contents.

It was thought that a comparison of the size of the particles of the sand in which the animal lived and of the particles of material in its intestine might reveal something about the feeding habits of the worm.

Accordingly a number of samples of sand were taken from the surface area of the beach where the animals were collected and the same number of samples from the sand about 25 cm below the surface. The samples were washed with distilled water (care being taken to allow very fine particles to settle before decanting the water above them) and dried at 103°C. Samples of the dried sand, usually weighing 70-100 g, were passed through a series of sieves shaken mechanically for 20 minutes. The material that remained in each sieve was then weighed and the result expressed as a percentage of the total weight of the sample.

After each visit to Morgat about 150 sipunculids were placed in aerated sea-water for 2 days and their faecal matter collected, carefully washed and dried at 103°C. Samples, usually weighing 50-70 g, were then passed through the same series of sieves for 20 minutes. The diameters (in mm) of the wire mesh of the sieves

were 0.40, 0.315, 0.25, 0.20, 0.160, 0.125, 0.100, 0.080, 0.063 and 0.050.

The results of the granulometric analysis of the surface sand, the deeper sand and the intestinal contents are shown in Table 1. They are the mean of 20 analyses in each case.

TABLE I
Granulometric composition of sand and intestinal contents

Size of particles (μ)	Surface sand (%)	Deeper sand (%)	Intestinal Contents %
> 400	1.13	1.85	1.52
400 - 315 315 - 250 250 - 200	$egin{array}{c} 1.11 \ 2.35 \ 6.09 \end{array}$	$egin{array}{c} 1.13 \ 2.32 \ 6.06 \end{array}$	$egin{array}{c} 1.42 \ 2.71 \ 7.29 \ \end{array}$
200 - 160 160 - 125	$14.25 \\ 33.07$	14.93 30.81	15.49 32.39
125 - 100 100 - 80 80 - 63	$24.74 \\ 15.98 \\ 1.15$	24.84 16.11 1.17	22.72 14.75
63 - 50 < 50	$0.15 \\ 0.16$	$egin{pmatrix} 1.17 \\ 0.22 \\ 0.22 \end{smallmatrix}$	$\begin{array}{c} 0.90 \\ 0.23 \\ 0.19 \end{array}$
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Amount of carbonate in the sand and the intestinal contents.

A series of experiments were conducted to determine the amount of carbonate (assumed to be calcium carbonate) in the sand and in the intestinal contents of the animal.

The method used was that employed by Lafon (1953). Samples of dried sand (usually weighing about 10 g) were poured into an excess of 10% hydrochloric acid and allowed to stand until effervescence had stopped. After 24 hours the supernatant liquid was decanted and the remaining sand washed twice with tap and twice with distilled water. The sand was then dried at 103°C, cooled and weighed. The loss of weight was taken to be the amount of calcium carbonate in the sand decomposed by the acid. Lafon (1953) showed that this method gave results which differ by less than 3% from those obtained by more elaborate methods of quantitative analysis.

Samples were collected from the surface and the deeper layers (about 25 cm) of sand. Dried faecal material was used for intestinal contents.

The mean percentage of carbonate in (a) the surface samples of sand (20 analyses) was found to be 51.7 ± 1.9 (b) the deeper sand (20 analyses) 52.4 ± 2.0 and (c) the faecal matter (15 analyses) 52.1 ± 1.8 .

Amount of nitrogen in the sand and the intestinal contents.

In order to find out how much protein was contained in the environment of the animals Keldjahl estimations for nitrogen were made on freshly collected samples of surface sand and deeper sand.

Dried samples weighing 4-5 g were digested with concentrated suppluric acid and catalyst. The ammonia liberated after treatment with alkali was bubbled through dilute acid and back-titrated with standard alkali.

The gut of 10 freshly collected specimens was ligatured in two places and the outer surface carefully washed with distilled water in order to remove any adhering proteins in solution from its walls. The canal was then cut and the intestinal contents freed in a vessel. The material was dried and weighed and the amount of nitrogen it contained determined. The results are shown in Table II.

TABLE II

Mean percentages of nitrogen and protein in samples

	surface sand (10 analyses)	deeper sand (10 analyses)	intestinal contents (9 analyses)
nitrogen	0.025 ± 0.008	0.030 ± 0.007	0.044 ± 0.014
protein (Nx6.25)	0.16 ± 0.05	0.19 ± 0.05	0.28 ± 9.09

Hydrogen ion concentration of the intestinal contents.

The results of the experiments on the determination of carbonate showed that the calcium carbonate in the sand ingested by *S. nudus* passed through the animal without solution. Experiments were therefore conducted to determine the pH of the intestinal contents. Electrometric and colorimetric (using indicator papers) methods were used. In the first case fluid was taken from the gut of animals that had been fasting for two days and on the second the intestinal contents of freshly collected animals was used. Attempts to trace the change of pH along the gut were not very successful. The results, however, are given for the anterior region of the gut (from the mouth to the beginning of the intestinal coils) and the posterior regions (the intestinal coils); ten determinations were made in each case.

TABLE III
pH of the intestinal contents

	electrometric	colorimetric
anterior region posterior region	7.7 ± 0.2 7.8 ± 0.3	7.8±0.3 7.9±0.3

Absorption of water from the alimentary canal.

had been collected the contents of the anterior region of the gut It was noticed that when specimens were opened just after they were usually more fluid than those of the posterior region. Experiments were therefore carried out to ascertain whether water was absorbed during the passage of food through the alimentary canal. The method used was that employed by Kermac (1955) to show the absorption of water from the gut of Arenicola marina. The body cavity of a number of worms was opened within 15 minutes of collecting the animals and the body fluid allowed to drain off. By means of small clips, the alimentary canal was ligatured in four places - (a) near the mouth - (b) at the beginning of the downward loop of the intestine (the region a - b comprising the foregut) -(c) at the bottom of the intestinal loop (b - c comprising the mid-gut) and - (d) near the anal aperture (c - d comprising the hind-gut). The numerous fastening strands connecting the alimentary canal to the body wall were then severed, the section of the gut between the ligatures cut and the section and its contents quickly dropped into small stoppered tubes that had been previously weighed. The tubes were stoppered tightly, returned to the laboratory, reweighed and heated to a constant weight at 103°C. Thus for each worm the percentage of water in the fore-, mid- and hind-gut was calculated.

One source of error of the method used is that the result gives the percentage of water contained in the tissue of the gut as well as the contents of the gut. As the gut wall is very thin and fairly uniform in thickness, it was considered that the error was small.

A series of experiments was carried out on five occasions, ten determinations being made each time. The results are given in Table IV.

TABLE IV

Percentage of water in the intestinal contents of S. nudus at different regions of the gut

Group (each consists of 10 animals)	fore-gut	mid-gut	hind-gut
1	49.1	40.5	33.3
2	55.2	47.4	34.7
3	55.0	44.6	37.8
4	53.1	44.2	40.9
5	47.1	42.7	35.7
Mean values (%)	51.9±3.2	44.8±2.4	$36.5\!\pm\!2.4$

DISCUSSION AND CONCLUSIONS

Specimens of *S. nudus* were found to be numerous on the sandy beach at Morgat during the late summer of 1960. They were collected at a level just above that of low water spring tide. To what extent the animals occur below the low water mark was not studied. Usually only adult specimens were found.

The sand in which the animals lived was very fine, 55% of the particles (by weight) having a diameter of $(100\text{-}160)~\mu$. No significant difference was found between the granulometric composition of the surface and deeper sand. The results also show that although the sand is fine it contained very little mud (less than 0.25%), particles of the latter being considered to have a diameter of $(5\text{-}50)~\mu$. The sand at Morgat contained a high percentage of carbonate (50-54%) and a low percentage of nitrogen (0.017-0.037%). These results are in general agreement with those obtained for the sands of lower Normandy by Lafon (1953) and for Heligoland by Krüger (1958).

The results show (a) that the granulometric composition of the sand and of the intestinal contents of the animal was almost the same and (b) that the percentage of carbonate in the sand and in the intestinal contents was almost the same. It appears, therefore, that the animal did not select either particles of sand or carbonate when it fed. Since large quantities of sand were always found in the gut it seems probable that the animal obtained at least some of its food from the organic matter contained in the sand.

The amount of nitrogen in the intestinal contents of nine animals was found to be about double that contained in the sand. This suggests that S. nudus obtained some of its food from the sea possibly by a ciliary-feeding process as mentioned by Chin and Wu (1950) and Tetry (1959 p. 834) or by filtering or selecting organic matter from the sand. Krüger (1958) found that A. marina was able to use sand to filter food particles from a current of water and Peebles and Fox (1933) found that Dendrostomum zostericolum was able to ingest gelatin particles from sea water. The results of the present investigation, however, do not justify either of such conclusions because it is not known how much protein was secreted by the animal in its digestive juices and in its intestinal mucus. It is possible that the flow of these fluids might account for the increased amount of nitrogen in the intestine.

Chin and Wu (1950), having recovered great numbers of diatoms from the intestinal contents of *S. nudus* (as it is found in Quamoy, China), concluded that the animal depended largely on diatoms for its food which it obtained by the movement of its tentacles or "by eating organic matter in the manner of an earthworm". On four different occasions a microscopic examination was made of (1) the contents of the anterior 3 cm of the gut of four freshly collected worms and (2) the surface sand at Morgat. Frustules of diatoms were found in all samples. Some of the diatoms in the gut looked as if they might have been alive when ingested but most were devoid of organic matter and possibly so when swallowed. Although *S. nudus* might have obtained some of its food from the diatoms it is difficult to conclude from the limited amount of information gathered whether diatoms were the principal source of food of the worms at Morgat.

Particles of carbonate contained in the ingested sand were not decomposed as they passed through the gut of *S. nudus*. The pH of the intestinal contents was found to be 7.5 - 8.1. This is in accord with the work of Enrique (1903) who found that the fluid in the gut *S. nudus* was alkaline.

Water was absorbed from the intestinal contents as they passed along the alimentary canal. Kermac (1955) also found that water was absorbed from the gut of A. marina. Sections of tissue stained with Alcian blue and mucicarmine showed that a considerable number of mucus secreting glands were present in the walls of the hind-gut and rectum of S. nudus. The mucus may help the less moist faecal material to be voided.

Acknowledgements

I wish to acknowledge the valuable and ready help given to me by the Director and staff of the Biological Station at Roscoff.

Summary

Specimens of S. nudus were found in considerable numbers in the sandy beach at Morgat during August and September, 1960. From a strip of beach $100~\mathrm{m} \times 10~\mathrm{m}$, sampled on two occasions, 4.6 ± 0.98 sipunculids per square metre were collected. The sand in which the animals lived was found to by very fine and to contain very little mud, a high percentage of carbonate and a low percentage of nitrogen. The granulometric composition of the particles and the percentage of carbonate in the intestinal contents and in the sand in which the animal lived were found to be almost the same. The amount of nitrogen in the intestinal contents was about twice that in the sand. It was difficult to assess the importance of the result because the amount of nitrogen secreted in the digestive juices and the intestinal mucus was not known. Some skeletons of diatoms were found in the intestinal contents of the anterior gut. Most of them were devoid of organic matter and were probably so when ingested. In the circumstances it is difficult to say how important a part diatoms played in the food of S. nudus at Morgat. The particles of carbonate contained in the ingested sand were not decomposed in the gut. The pH of the intestinal contents was found to be 7.5 - 8.2. Water was absorbed from the intestinal contents as they passed along the alimentary canal.

Zusammenfassung

Exemplare von Sipunculus nudus wurden in grosser Zahl im Jahre 1960 während der Monate August und September am Sandstrand von Morgat gefunden. Auf einem Strandstreifen von 100 m auf 10 m, auf dem zwei mal gesammelt wurde, fanden sich 4,6 ± 0,98 Sipunculiden pro Quadratmeter. Der Sand, in welchem die Tiere lebten, war sehr fein und enthielt wenig Schlamm, einen hohen Gehalt an Karbonaten (an Kohlenstoff?) und einen niedrigen Gehalt an Stisckstoff. Die Verteilung der Korngrösse der Teilchen und der Gehalt an Karbonaten im Darminhalt und im Sande, in dem die Tiere lebten, waren nicht verschieden. Der Stickstoffgehalt im Darminhalt war ungefähr zwei mal grössen als im Sande. Es war schwierig, die Bedeutung dieses Resultates festzusetzen, da die in den Verdauungssäften und in der Darmschleimhaut ausgeschiedene Stickstoffmenge nicht bekannt war. Im vorderen Darmabschnitt wurden im Darminhalt einige Diatomeenskelette gefunden. Sie enthielten meistens kein organisches Material und dies war wahrscheinlich schon so bei der Einverleibung. Unter diesen Umständen ist es schwierig zu sagen, welchen Anteil die Diatomeen in der Ernährung von S. nudus in Morgat ausmachen. Die im aufgenommenen Sand enthaltenen Karbonatteilchen wurden nich aufgespalten im Darmtrakt, Das pH des Darminhaltes schwankte zwischen 7,5 und 8,2 Während der Darminhalt den Darmtrakt durchläuft, wird Wasser aufgenommen.

REFERENCES

- CHIN, T.G. and WU, C.S., 1950. Diatoms in the intestines of Amoy Sipunculida (Annelida: Gephyrea). Ling. Sc. J., 23, pp. 43-51.
- ENRIQUES, P., 1903. I corpi pigmentati del Sipunculus nudus. Arch. Zool. Ital., 1, pp. 253-285.
- KERMAC, D.M., 1955. The anatomy and physiology of the gut of the polychaete Arenicola marina iL.). Proc. Zool. Soc. Lond., 125 (2), pp. 347-381.
- KRÜGER, F., 1958. Zur Ernährungsphysiologie von Arenicolo marina (L.). Verh. deutsch. Zool. Ges. Frankfurt, pp. 115-120.
- LAFON, M., 1953. Recherches sur les sables côtiers de la Basse-Normandie et sur quelques conditions de leur peuplement zoologique. Ann. Inst. Océan., 28 (3), pp. 113-161.
- PEEBLES, F. and Fox, D.L., 1933. The structure, function and general reactions of the marine sipunculid worm, Dendrostoma zostericola. Bull. Scripps Inst. Oceanogr. Tech., 3 (9), pp. 201-24.
- TETRY, A., 1959. « Classe des Sipunculiens » in Traité de Zoologie (ed. P. Grassé), V (1), pp. 785-854.